

WHAT IS CLAIMED IS:

1. A method for detecting the presence or absence of VZV in a biological sample
5 from an individual, said method comprising:

performing at least one cycling step, wherein a cycling step comprises an
amplifying step and a hybridizing step, wherein said amplifying step comprises amplifying a
portion of a VZV gene 28 nucleic acid molecule from said biological sample using a pair of gene
28 primers, thereby producing a gene 28 amplification product, wherein said hybridizing step
10 comprises hybridizing a pair of gene 28 probes to said gene 28 amplification product, wherein
the members of said pair of gene 28 probes hybridize within no more than five nucleotides of
each other, wherein a first gene 28 probe of said pair of gene 28 probes is labeled with a donor
fluorescent moiety and said second gene 28 probe of said pair of gene 28 probes is labeled with a
corresponding acceptor fluorescent moiety; and

detecting the presence or absence of fluorescence resonance energy transfer
(FRET) between said donor fluorescent moiety of said first gene 28 probe and said acceptor
fluorescent moiety of said second gene 28 probe upon hybridization of said pair of gene 28
probes to said amplification product,

wherein the presence of FRET is indicative of the presence of VZV in said
20 biological sample, and wherein the absence of FRET is indicative of the absence of VZV in said
biological sample.

2. The method of claim 1, wherein said pair of gene 28 primers comprises a first
gene 28 primer and a second gene 28 primer, wherein said first gene 28 primer comprises the
25 sequence

5'-GAC AAT ATC ATA TAC ATG GAA TGT G-3' (SEQ ID NO:1), and wherein said
second gene 28 primer comprises the sequence

5'-GCG GTA GTA ACA GAG AAT TTC TT-3' (SEQ ID NO:2).

3. The method of claim 1, wherein said first gene 28 probe comprises the sequence

5'-CGA AAA TCC AGA ATC GGA ACT TCT T-3' (SEQ ID NO:3), and wherein said second gene 28 probe comprises the sequence

5'-CCA TTA CAG TAA ACT TTA GGC GGT C-3' (SEQ ID NO:4).

5 4. The method of claim 1, wherein the members of said pair of gene 28 probes hybridize within no more than two nucleotides of each other.

5 5. The method of claim 1, wherein the members of said pair of gene 28 probes hybridize within no more than one nucleotide of each other.

10 7. The method of claim 1, wherein said donor fluorescent moiety is fluorescein.

10 8. The method of claim 1, wherein said corresponding acceptor fluorescent moiety is selected from the group consisting of LC-Red 640, LC-Red 705, Cy5, and Cy5.5.

10 9. The method of claim 1, wherein said detecting step comprises exciting said biological sample at a wavelength absorbed by said donor fluorescent moiety and visualizing and/or measuring the wavelength emitted by said acceptor fluorescent moiety.

20 10. The method of claim 1, wherein said detecting comprises quantitating said FRET.

20 11. The method of claim 1, wherein said detecting step is performed after each cycling step.

25 12. The method of claim 1, wherein said detecting step is performed in real time.

30 13. The method of claim 1, further comprising determining the melting temperature between one or both of said gene 28 probe(s) and said gene 28 amplification product, wherein said melting temperature confirms said presence or said absence of said VZV.

14. The method of claim 1, wherein the presence of said FRET within 50 cycling steps is indicative of the presence of a VZV infection in said individual.

15. The method of claim 1, wherein the presence of said FRET within 40 cycling steps is indicative of the presence of a VZV infection in said individual.

16. The method of claim 1, wherein the presence of said FRET within 30 cycling steps is indicative of the presence of a VZV infection in said individual.

17. The method of claim 1, further comprising: preventing amplification of a contaminant nucleic acid.

18. The method of claim 17, wherein said preventing comprises performing said amplifying step in the presence of uracil.

19. The method of claim 18, wherein said preventing further comprises treating said biological sample with uracil-DNA glycosylase prior to a first amplifying step.

20. The method of claim 1, wherein said biological sample is selected from the group consisting of dermal swabs, cerebrospinal fluid, ganglionic tissue, brain tissue, ocular fluid, blood, sputum, bronchio-alveolar lavage, bronchial aspirates, lung tissue, and urine.

21. The method of claim 1, further comprising:
performing at least one cycling step, wherein said cycling step comprises an amplifying step and a hybridizing step, wherein said amplifying step comprises amplifying a portion of a VZV gene 29 nucleic acid molecule from said biological sample using a pair of gene 29 primers, thereby producing a gene 29 amplification product, wherein said hybridizing step comprises hybridizing a pair of gene 29 probes to said gene 29 amplification product, wherein the members of said pair of gene 29 probes hybridize within no more than five nucleotides of each other, wherein a first gene 29 probe of said pair of gene 29 probes is labeled with a donor

fluorescent moiety and said second gene 29 probe of said pair of gene 29 probes is labeled with a corresponding acceptor fluorescent moiety; and

detecting the presence or absence of FRET between said donor fluorescent moiety of said first gene 29 probe and said acceptor fluorescent moiety of said second gene 29 probe upon hybridization of said pair of gene 29 probes to said amplification product.

22. The method of claim 21, wherein said pair of gene 29 primers comprises a first gene 29 primer and a second gene 29 primer, wherein said first gene 29 primer comprises the sequence

5'-TGT CCT AGA GGA GGT TTT ATC TG-3' (SEQ ID NO:5),

and wherein said second gene 29 primer comprises the sequence

5'-CAT CGT CTG TAA GAC TTA ACC AG-3' (SEQ ID NO:6).

23. The method of claim 21, wherein said first gene 29 probe comprises the sequence

5'-GGG AAA TCG AGA AAC CAC CCT ATC CGA C-3' (SEQ ID NO:7), and wherein

said second gene 29 probe comprises the sequence

5'-AAG TTC GCG GTA TAA TTG TCA GTG GCG-3' (SEQ ID NO:8).

24. The method of claim 1, wherein said cycling step is performed on a control sample.

25. The method of claim 24, wherein said control sample comprises said portion of said VZV gene 28 nucleic acid molecule.

26. The method of claim 1, wherein said cycling step uses a pair of control primers and a pair of control probes, wherein said control primers and said control probes are other than said gene 28 primers and gene 28 probes, wherein said amplifying step produces a control amplification product, wherein said control probes hybridize to said control amplification product.

27. A method for detecting the presence or absence of VZV in a biological sample from an individual, said method comprising:

performing at least one cycling step, wherein a cycling step comprises an amplifying step and a hybridizing step, wherein said amplifying step comprises amplifying a portion of a VZV gene 29 nucleic acid molecule from said biological sample using a pair of gene 29 primers, thereby producing a gene 29 amplification product, wherein said hybridizing step comprises hybridizing a pair of gene 29 probes to said gene 29 amplification product, wherein the members of said pair of gene 29 probes hybridize within no more than five nucleotides of each other, wherein a first gene 29 probe of said pair of gene 29 probes is labeled with a donor fluorescent moiety and said second gene 29 probe of said pair of gene 29 probes is labeled with a corresponding acceptor fluorescent moiety; and

detecting the presence or absence of fluorescence resonance energy transfer (FRET) between said donor fluorescent moiety of said first gene 29 probe and said acceptor fluorescent moiety of said second gene 29 probe upon hybridization of said pair of gene 29 probes to said amplification product,

wherein the presence of FRET is indicative of the presence of VZV in said biological sample, and wherein the absence of FRET is indicative of the absence of VZV in said biological sample.

28. An article of manufacture, comprising:

a pair of gene 28 primers;

a pair of gene 28 probes; and

a donor fluorescent moiety and a corresponding acceptor fluorescent moiety.

29. The article of manufacture of claim 28, wherein said pair of gene 28 primers comprise a first gene 28 primer and a second gene 28 primer, wherein said first gene 28 primer comprises the sequence

5'-GAC AAT ATC ATA TAC ATG GAA TGT G-3' (SEQ ID NO:1),

and wherein said second gene 28 primer comprises the sequence

5'-GCG GTA GTA ACA GAG AAT TTC TT-3' (SEQ ID NO:2).

30. The article of manufacture of claim 28, wherein said pair of gene 28 probes comprises a first gene 28 probe and a second gene 28 probe, wherein said first gene 28 probe comprises the sequence

5'-CGA AAA TCC AGA ATC GGA ACT TCT T-3' (SEQ ID NO:3),

5 and wherein said second gene 28 probe comprises the sequence

5'-CCA TTA CAG TAA ACT TTA GGC GGT C-3' (SEQ ID NO:4).

31. The article of manufacture of claim 28, wherein said first gene 28 probe is labeled with said donor fluorescent moiety and wherein said second gene 28 probe is labeled with said
10 corresponding acceptor fluorescent moiety.

32. The article of manufacture of claim 28, further comprising a package insert having instructions thereon for using said pair of gene 28 primers and said pair of gene 28 probes to detect the presence or absence of VZV in a biological sample.

33. An article of manufacture, comprising:
a pair of gene 29 primers;
a pair of gene 29 probes; and
a donor fluorescent moiety and a corresponding acceptor fluorescent moiety.

34. The article of manufacture of claim 33, wherein said pair of gene 29 primers comprises a first gene 29 primer and a second gene 29 primer, wherein said first gene 29 primer comprises the sequence

5'-TGT CCT AGA GGA GGT TTT ATC TG-3' (SEQ ID NO:5),

25 and wherein said second gene 29 primer comprises the sequence

5'-CAT CGT CTG TAA GAC TTA ACC AG-3' (SEQ ID NO:6).

35. The article of manufacture of claim 33, wherein said pair of gene 29 probes comprises a first gene 29 probe and a second gene 29 probe, wherein said first gene 29 probe
30 comprises the sequence

5'-GGG AAA TCG AGA AAC CAC CCT ATC CGA C-3' (SEQ ID NO:7),

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